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     (FILE 'HOME' ENTERED AT 20:07:36 ON 27 SEP 2003)
     FILE 'CA' ENTERED AT 20:07:46 ON 27 SEP 2003
L1 107781 S (CELL OR CELLULAR)(4A)(PREPAR? OR EXAMIN? OR STAIN?)
L2   8484 S L1 AND(FILTER? OR FILTRAT? OR SEPARAT?)
L3    173 S L2 AND(MICROSCOPE OR SLIDE)
L4   1451 S L2 AND(AGITAT? OR MIX?)
L5     27 S L3 AND L4
     FILE 'BIOSIS' ENTERED AT 20:27:07 ON 27 SEP 2003
L6     16 S L5
     FILE 'MEDLINE' ENTERED AT 20:29:27 ON 27 SEP 2003
L7     20 S L5
L8    556 S L4
L9    421 S L8 NOT FLOW CYTOM? NOT PY>1999
L10     7 S L9 AND AUTOMAT?
L11   178 S L9 AND(CYTOLO? OR HISTO?)
L12    29 S L11 AND(LAYER OR FILM OR SUSPEN?)(2A)(CELL OR CELLULAR OR PARTICLE)
     FILE 'BIOSIS' ENTERED AT 20:34:28 ON 27 SEP 2003
L13     7 S L12
L14     5 S L10
     FILE 'CA, BIOSIS, MEDLINE' ENTERED AT 20:37:04 ON 27 SEP 2003
L15    81 DUP REM L5 L6 L13 L14 L7 L12 L10 (30 DUPLICATES REMOVED)

=> d bib,ab l15 1-81

L15  ANSWER 8 OF 81  CA  COPYRIGHT 2003 ACS on STN
AN   132:20777  CA
TI   Method for staining biological specimens by combining stable reagents on a
     microscope slide to make an unstable staining solution
IN   Mehta, Parula; Graham, Marshal; Pomerantz, Anlouise
PA   Ventana Medical Systems, Inc., USA
SO   PCT Int. Appl., 28 pp.
PI   WO 9963342        A1   19991209       WO 1999-US12263  19990602
PRAI US 1998-87673P    P    19980602
AB   The present invention relates to automated methods for staining biolocal
     materials on a slide comprising the use of component histochem. solns. mixed
     directly on a biol. sample of interest.  The method comprises providing at
     least two stable solns. that together comprise an unstable staining soln.,
     sequentially delivering the stable solns. to a biol. sample of interst on a
     surface, and mixing the stable solns. directly on the biol. material of
     interest to effectuate staining of the material.  An automated protocol was
     used to stain Aspergillus cryptococcus samples by the Grocott's method for
     fungi staining.  The silver nitrate and the methanamine-borax solns. were
     added sep.

L15  ANSWER 20 OF 81     MEDLINE on STN
AN   96128422     MEDLINE
TI   Efficiency of various dissociation methods for the preparation of thyroid
     single cell suspensions.
AU   Frohlich E; Wahl R; Reutter K
CS   Department of Anatomy, Eberhard-Karls-University Tubingen.
SO   EXPERIMENTAL AND CLINICAL ENDOCRINOLOGY AND DIABETES, (1995) 103 (5) 308-16.
AB   For comparison of the physiological potential of single thyroid cells versus
     cells integrated into follicles it would be ideal to work with suspensions
     consisting exclusively of single cells instead of a mixture of single cells
     and follicle fragments.  In this study, various techniques for the isolation
     of single cells have been tested for their effect on cell viability, the
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ultrastructure of the isolated cells, the percentage of single cells and the ability of these cells to form follicles in culture. In addition, the cells were characterized for the preservation of their morphology and the ability to respond to TSH by comparing their immunocytochemical staining pattern with anti-vimentin and anti-ras p21 antibody to that of the intact thyroid tissue. Dispase treatment of thyroid tissues alone produced suspensions with a relatively small proportion of single cells. These **cells stained** with anti-vimentin and anti-ras p21 antibody to a similar percentage as thyroid cells in the intact gland. A combination of dispase treatment with either **filtration** or trypsin treatment severely compromised the viability of the cells. A high proportion of single cells with a good viability could be obtained either by centrifugation of dispase treated tissues or by culturing of dispase treated tissues as monolayers and subsequent detachment from the culture vessels with trypsin. Whereas the immunological staining with anti-vimentin and anti-ras oncogene antibody in the centrifuged cells resembled that of intact tissue, cells cultured as monolayers reacted differently. The differences in the immunological staining were still observed when the cells which had been grown as monolayers were stimulated with TSH. Differential centrifugation appeared to be the ideal method for the isolation of unaltered and viable single cells but is a rather laborious method to obtain larger amounts of single thyroid cells.

L15 ANSWER 25 OF 81 MEDLINE on STN
AN 92328806 MEDLINE
TI Activin A increases cytosolic free calcium concentration in rat pituitary somatotropes.
AU Tasaka K; Kasahara K; Masumoto N; Mizuki J; Kurachi H; Miyake A; Tanizawa O
CS Department of Obstetrics and Gynecology, Osaka University Medical School, Japan.
SO BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, (1992 Jun 30) 185 (3) 974-80.
AB The effect of activin A on the cytosolic free calcium concentration ([Ca2+]i) in normal rat pituitary **cells** was **examined** using a calcium sensitive fluorescent dye, indo 1 AM, and a digital imaging fluorescent **microscope** system. The cells showing an increase in [Ca2+]i in response to activin A were then characterized by comparison with cells responding to growth hormone releasing hormone (GRH), thyrotropin releasing hormone (TRH), corticotropin releasing hormone (CRH), and gonadotropin releasing hormone (GnRH) in monolayer cultures of normal rat pituitary cells. Activin A increased [Ca2+]i in some cells in a **mixed** population of normal rat pituitary cells. The cells that responded to activin A also responded to GRH. Most of these cells were not affected by other tropic hormones (CRH, TRH, and GnRH), but a few cells responded to both GRH and TRH. None of the activin A-responding cells responded to CRH or GnRH, and none of the CRH- or GnRH-responding cells responded to activin A. In a preparation of somatotropes purified 80-90% by fluorescence-activated cell sorting, activin A increased [Ca2+]i in 30% of the cells that shows a [Ca2+]i-response to GRH. These findings suggest direct involvement of somatotropes in activin A-induced biological events in the rat pituitary gland.

L15 ANSWER 28 OF 81 MEDLINE on STN
AN 91253332 MEDLINE
TI Large-volume cytocentrifuge for processing alcohol-fixed cytologic specimens. Application in urinary cytology.
AU Toivonen T; Hasto A L
CS Department of Pathology, Maria Hospital, Helsinki, Finland.
ACTA CYTOLOGICA, (1991 May-Jun) 35 (3) 269-72.
The use of a large-volume cytocentrifuge for the routine processing of urine

specimens was investigated. Most specimens arrived in the laboratory **mixed** with 50% ethanol. Polyethylene glycol was added to the fixative supplied to the clinicians or to the cell suspension received in the laboratory. The **slides** used in the cytocentrifuge were coated with gelatin or poly-L-lysine to minimize cell loss. The resulting monolayers of cells, with occasional true tissue fragments, were stained by the Papanicolaou method. When indicated, immunocytochemical and histochemical stains were used. The technique gave good morphologic details, good **cell** yield and consistent **staining** quality. This method has also been applied to other cytologic specimens, such as serous fluids and fine needle aspirates, with good results. The method saves the time of cytotechnologists, and **slides** prepared by this technique are suitable for automatic staining.

L15 ANSWER 31 OF 81 MEDLINE on STN
AN 91104000 MEDLINE
TI A multiwell cell settling and adherence chamber for morphology and differential counting.
AU Leonard E J; Yoshimura T; Skeel A; Goodwin R
CS Laboratory of Immunobiology, National Cancer Institute, FCRDC, Frederick, MD 21702.
SO BIOTECHNIQUES, (1990 Dec) 9 (6) 684, 686, 688-9.
AB A simple multiwell chamber is described that can be used to **prepare** randomly distributed **cells** on a **microscope slide**, suitable for morphological identification and differential counting. To the eight wells of the chamber are added 50-microliter volumes of cell suspension at concentrations of 10(3)-10(6) cells/ml. As the cells settle, fluid is slowly wicked away by a damp **filter** paper sandwiched between the **microscope slide** and the acrylic top plate of the multiwell chamber. Within 20-40 minutes, the cell monolayers on the **slide** are completely dry. The combined settling and bulk fluid removal results in a distribution of adherent cells that are sufficiently spread to exhibit excellent morphology after staining. If the chamber is centrifuged for 30 seconds at 50x g immediately after addition of cells, recovery of cells in the monolayer is virtually 100%, and as few as 50 input cells per 50 microliters can be detected. Agreement between predicted and observed differential counts of cell **mixtures** indicates that cells in the monolayer were distributed randomly.

L15 ANSWER 41 OF 81 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1988:466758 BIOSIS
TI BRONCHOALVEOLAR LAVAGE CELL DIFFERENTIAL ON **MICROSCOPE** GLASS COVER A SIMPLE AND ACCURATE TECHNIQUE.
AU LAVIOLETTE M; CARREAU M; COULOMBE R
CS CENT. DE PNEUMOLOGIE, HOPITAL LAVAL 2725 CHEMIN STE-FOY, STE-FOY, QUEBEC G1V 4G5 CANADA.
SO AM REV RESPIR DIS, (1988) 138 (2), 451-457.
AB We describe a quick and easy technique to perform cell differentials on bronchoalveolar lavage: the **microscope** glass cover. Lavage fluids of 72 subjects were analyzed by 3 techniques: glass cover, **filter**, and cytocentrifuge preparations. Seventy-seven other lavages were analyzed by glass cover and cytocentrifuge preparations alone. Data for the 72 subjects studied by all 3 techniques showed that the cell counts on glass cover and **filter** preparations were similar, e.g., lymphocytes, 19.2 (range, 0.5 to 94%) and 20.9% (range, 3 to 95%), respectively (Spearman's correlation coefficient, 0.98). However, on cytocentrifuge preparations, lymphocyte counts were lower (8.3%; range, zero to 87%) and macrophage counts were higher ($p < 0.005$). Comparison of glass cover and cytocentrifuge preparation **mixtures** with varying amounts (20 to 80%) of purified blood leukocytes labeled with 51Cr (≥ 72% lymphocytes) showed that a significant amount of

radioactive cells was lost during the cytocentrifuge technique in contrast to the glass cover technique. Because neutrophils represented a low proportion of lavage cells, we also evaluated cell suspensions with known neutrophils contents (10 to 70%); we found no difference in neutrophil counts obtained with the 3 techniques. Lavage data analysis of 40 young nonsmoking volunteers showed that glass cover lymphocyte count was also higher than counts on cytocentrifuge preparations: 16.5% (range, 3 to 45%) and 8.2% (range, 2.5 to 35%), respectively. In this group, the distribution of glass cover lymphocyte percentage was normal ($p = 0.21$, $\chi 2$ test), and the one-tailed 95% confidence interval was 18.6 to 34.7% (mean plus 1.65 standard deviation). This study shows that the glass cover **preparation**, besides giving an accurate **cell** count, is technically advantageous and preserves cell structure. Therefore, the glass cover, is technique should be considered as a valuable alternative to the **filter** and cytocentrifuge techniques for the determination of lavage cell differentials.

L15 ANSWER 50 OF 81 MEDLINE on STN
AN 85020898 MEDLINE
TI Accurate quantification of cells recovered by bronchoalveolar lavage.
AU Saltini C; Hance A J; Ferrans V J; Basset F; Bitterman P B; Crystal R G
SO AMERICAN REVIEW OF RESPIRATORY DISEASE, (1984 Oct) 130 (4) 650-8.
AB Quantification of the differential cell count and total number of cells recovered from the lower respiratory tract by bronchoalveolar lavage is a valuable technique for evaluating the alveolitis of patients with inflammatory disorders of the lower respiratory tract. The most commonly used technique for the evaluation of cells recovered by lavage has been to concentrate cells by centrifugation and then to determine total cell number using a hemocytometer and differential cell count from a Wright-Glemsa-stained cytocentrifuge preparation. However, we have noted that the percentage of small cells present in the original **cell suspension** recovered by lavage is greater than the percentage of lymphocytes identified on cytocentrifuge preparations. Therefore, we developed procedures for determining differential cell counts on lavage cells collected on Millipore **filters** and stained with hematoxylin-eosin (**filter** preparations) and compared the results of differential **cell** counts performed on **filter preparations** with those obtained using cytocentrifuge preparations. When cells recovered by lavage were collected on **filter preparations**, accurate differential **cell** counts were obtained, as confirmed by performing differential cell counts on cell **mixtures** of known composition, and by comparing differential **cell** counts obtained using **filter preparations** stained with hematoxylin-eosin with those obtained using **filter** preparations stained with a peroxidase cytochemical stain. The morphology of **cells** displayed on **filter preparations** was excellent, and interobserver variability in quantitating cell types recovered by lavage was less than 3%.(ABSTRACT TRUNCATED AT 250 WORDS)

L15 ANSWER 52 OF 81 MEDLINE on STN
AN 83303026 MEDLINE
TI Measurement of human endothelial cells in whole blood.
AU Takahashi H; Harker L A
SO THROMBOSIS RESEARCH, (1983 Jul 1) 31 (1) 1-12.
AB To evaluate the usefulness of measuring circulating endothelial cells as an in vivo indicator for vascular injury, studies were performed in vitro using cultured human endothelial cells and Factor VIII-related antigen (VIIIR:Ag) as the specific cell marker. Cultured human endothelial cells **mixed** with normal whole blood were processed by Ficoll-Hypaque gradient sedimentation to remove red cells and granulocytes, followed by pellet formation of the mononuclear supernatant fraction to **separate** the bulk of platelets. **Cell**

films, **prepared** from a measured volume of the final **cell suspension**, were **stained** with fluorescent antibody against VIII R:Ag, and positively **stained cells** were counted under fluorescence microscopy. The final recovery of endothelial cells was 64.8 +/- 19.3% (n = 41), and the minimal detectable concentration of endothelial cells was about 0.02 cells/microliter whole blood. No endothelial cells were detected in the blood of normal individuals.

L15 ANSWER 59 OF 81 MEDLINE on STN
AN 82031828 MEDLINE
TI Cell sorter immunofluorescence detection of human erythrocytes labelled in suspension with antibodies specific for hemoglobin S and C.
AU Bigbee W L; Branscomb E W; Weintraub H B; Papayannopoulou T; Stamatoyannopoulos G
SO JOURNAL OF IMMUNOLOGICAL METHODS, (1981) 45 (2) 117-27.
AB We have developed an immunochemical method for labeling human red blood cells in suspension with hemoglobin-specific antibodies. A membrane permeable cross-linking reagent, dimethyl suberimidate, is used to covalently bind, in situ, a fraction of the intracellular hemoglobin to integral membrane proteins. Hypotonic lysis and washing of the cells removes the unbound hemoglobin resulting in red blood cell ghosts which are permeable to macromolecules. Fluorescein-labeled antibodies for the hemoglobin variants S and C bind specifically to hemoglobin AS and AC ghosts, respectively, and not to normal hemoglobin AA ghosts. This technique can be used to **prepare** ghost suspensions for **cell** sorter analysis in which large numbers (10(9)--10(10)) of normal ghosts can be rapidly screened for the presence of rare anti-hemoglobin S and anti-hemoglobin C binding ghosts. In reconstruction experiments using **mixtures** of AS and AA cells and anti-hemoglobin S, AS ghosts as rare as 3 X 10(-5) were quantitatively recovered. Fluorescence artifacts prevented direct enumeration of AS ghosts at lower frequencies, but a two-step flow sorting-fluorescence **microscope** visual scanning procedure allows semiquantitative detection of anti-hemoglobin S-labeled ghosts as low as 10(--7). This method can be used for rapidly screening blood samples from individuals of normal hemoglobin A genotype for the presence of rare anti-hemoglobin S and anti-hemoglobin C binding ghosts.

L15 ANSWER 67 OF 81 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1977:180847 BIOSIS
TI OPTICAL PROPERTIES OF NORMAL AND INJURED CELLS APPLICATION OF CYTOGRAPHIC ANALYSIS OF CELL VIABILITY AND VOLUME STUDIES.
AU PENTTILA A; MCDOWELL E M; TRUMP B F
SO J HISTOCHEM CYTOCHEM, (1977) 25 (1), 9-20.
AB A cell spectrophotometer (Cytograf Model 6300 A, Bio/Physics Systems, Inc.) was tested in a cytotoxic assay using Ehrlich ascites [mouse] tumor cells as a model system. Several cellular conditions associated with volume expansion, **staining** of **cellular** components and fixation of cells were applied and the magnitude of the scattering and extinction signals were tested in these diverse cellular conditions. The magnitude of the scattering pulse of a cell spectrophotometer was greatly dependent on the **staining** color and intensity of **cellular** components with vital dyes or following OsO4 fixation. When the absorption wavelength of the vital dye was close to the wavelength used in the cell spectrophotometer (about 630 nm), dead **stained** and living nonstained **cell** populations were completely **separated** from each other. The magnitude of the extinction pulse was greatly dependent on the state (normal, injured **cells**) and **staining** intensity (vital dye **staining**, Os fixation) of **cellular** components. The magnitude of the extinction pulse was reduced from that in normal cells when cells were treated with p-

chloromercuribenzene sulfonic acid or in a hypotonic solution that caused a marked volume expansion of injured cells. When cells were fixed with a **mixture** containing glutaraldehyde and OsO4 the cellular components of normal and injured cells turned black and in these conditions the magnitude of the extinction signal was in a linear correlation to the cross-sectional area of cells. In the present study, the cell spectrophotometer proved to be an efficient method for estimation of cellular viability, based on different scattering properties of cells, offering the advantages of high speed and precision. Demonstration of the use of variety of vital dyes having diverse extinction properties with capabilities to differentiate between living and dead cells has indicated the potential use of the cell spectrophotometer in cytotoxic assay. Modification of the magnitude of the extinction signal in the cell spectrophotometer also shows great promise for accurate **automated** size determination of both normal and injured cells. Previously, determination of the size of injured cells was beset with methodologic errors.

L15 ANSWER 73 OF 81 CA COPYRIGHT 2003 ACS on STN
AN 64:70149 CA
OREF 64:13174d-f
TI Membrane properties of living mammalian cells as studied by enzymic hydrolysis of fluorogenic esters
AU Rotman, Boris; Papermaster, Ben W.
CS Syntex Inst. of Mol. Biol., Stanford Ind. Park, Palo Alto, CA
SO Proceedings of the National Academy of Sciences of the United States of America (1966), 55(1), 134-41
AB Fluorescein diacetate was **mixed** on a **microscope slide** with a cell suspension of cultured mouse lymphoma. An immediate bright green fluorescence appeared inside the **cell** when **examd.** under dark field with appropriate **filters**. This reaction was called fluorochromasia. Aspects of fluorochromasia were defined exptl. All cultivated mammalian lines examd. exhibited fluorochromasia, whereas a heterogeneity of fluorochromatic reactions occurred in vivo. Quant. studies of fluorochromasia with fluorescein analogs, at the population and single cell level, were consistent with a proposed model which stated that the cell membrane was permeable to the substrate, a non-polar compd., and less permeable to the polar product, fluorescein.

Q 11, 12, 26

=> log y
STN INTERNATIONAL LOGOFF AT 20:37:51 ON 27 SEP 2003